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Microbiological and Chemical Changes in Silage<sup>1/</sup>

C. W. Langston, Herbert Irvin<sup>2/</sup>, C. H. Gordon, Cecelia Bouma,  
H. G. Wiseman, C. G. Melin and L. A. Moore,

Dairy Cattle Research Branch

and

J. R. McCalmont

Agricultural Engineering Research Branch

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The problems involved in making grass silage are varied and many. Advances in our techniques have been slowed by inadequate definitions of silage quality and by emphasis that have been placed on certain practical aspects of silage making. Repeated ensiling of crops without preservatives under apparently similar conditions results in unexplained successes and failures. Relatively high moisture forages have most frequently shown this variability in efficiency of preservation and silage quality. The variability in results obtained indicates a lack of understanding of the basic factors and processes controlling silage quality. Because of the emphasis that has been placed on grassland farming and the importance of silage making in our present agricultural system, the need of fundamental bacteriological and chemical studies on silages is apparent.

#### EXPERIMENTAL PROCEDURES AND RESULTS

The experimental procedures were designed to provide a range in silage quality. By providing variations and at the same time studying the bacteriological and chemical changes involved it was felt that some of the underlying mechanisms affecting silage quality might emerge.

Silages were prepared from first, second and third cutting orchardgrass and alfalfa. The forages were ensiled in 4' x 8' experimental silos. The treatments were basically variations in which atmospheric oxygen was included or excluded (table 1).

Samples were taken from the silos at different stages of fermentation for bacteriological and chemical analysis. Bacteriological analysis included "total anaerobic counts", lactic acid bacteria counts and anaerobic spore

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1/ Paper presented at the annual meeting of the American Dairy Science Association, June 17-19, 1958 at North Carolina State College, Raleigh, North Carolina.

2/ Present address - Davis and Elkins College, Elkins, West Virginia.

counts. Chemical analysis included pH, crude protein, ether extract, nitrogen free extract, ash, ammonia nitrogen, reducing sugars and organic acids.

The silages were divided into good and poor quality on the basis of pH, ammonia nitrogen, lactic acid, butyric acid and spore counts. No single criterion was satisfactory for classification purposes, and judgement in evaluating several criteria which have been shown to be associated with good or poor silages was involved in classifying the silages. Characteristics of the good and poor silages are given in table 2. It was usually true that the forages with the more radical treatment in regard to atmospheric oxygen made the poorer quality silages. Sixteen of the thirty silages studied were classified as good, ten poor, and four intermediate quality.

The average bacterial counts for good and poor quality orchardgrass and alfalfa silages are given in figure 1. It is apparent that with the exception of the zero day figures, the same general pattern of increase and decrease in numbers of bacteria were similar throughout the fermentation in good and poor quality silages. This indicated that similar organisms were being cultured from the roll tubes ("total anaerobic counts") and the plates (lactic acid bacteria counts). The zero day counts also revealed that the initial numbers of lactic acid bacteria had little bearing on the final quality of the silages. The primary difference in quality could be correlated well with the increase in sporeforming anaerobes. In the orchardgrass silages the sporeformers began their rise at 2 days and increased to 13,000,000 per gram of silage at 62 days. In the alfalfa silages no significant increase was found until the 41-day fermentation period, and the numbers that appeared were much lower than those found in the orchardgrass silages. This is not in agreement with the conventional theory that legumes are more difficult to ensile than grasses. It was also true that first cuttings of both orchardgrass and alfalfa were superior in quality to subsequent cuttings.

Average amounts of organic acids in good and poor quality orchardgrass and alfalfa silages are shown in figures 2 and 3. The good quality silages (figure 2) showed substantial increases in lactic acid throughout the fermentation process to about 9% at 20-62 days. Acetic acid increased to about 2% along with small amounts of succinic acid.

The poor quality silages (figure 3) showed the usual increase of lactic acid up to the 5-8 day fermentation period. At this same period butyric acid appeared and increased at the expense of lactic acid. Acetic acid increased normally along with small amounts of propionic, formic and succinic acids.

About 4,000 strains of bacteria were picked from highest dilutions of silages. The organisms were representative of different quality silages, various stages of fermentation and different cuttings. The organisms were grouped on the basis of tests commonly used in characterizing lactic acid bacteria. Representative strains from the above groupings were then picked for detailed study. This work is currently being conducted. Upon completion,

we should have a better knowledge of the action of microorganisms at different stages of fermentation and in different quality silages.

#### SUMMARY

Orchardgrass and alfalfa forages were subjected to various treatments of air inclusion or exclusion to provide silages of different quality. The forages were prepared in this manner to study the bacteriological and chemical processes involved in good and poor silages.

The results of the study showed that by varying the inclusion or exclusion of air in forages, different quality silages could be obtained. The good quality silages had pH values of 4.8 or below, low ammonia nitrogen, some contained small amounts of butyric acid, but usually none was found, practically no sporeforming anaerobes were observed and all contained considerable amounts of lactic acid. A reversal of the above was noted for the poor quality silages.

First cutting forages were superior in quality to subsequent cuttings. Alfalfa silages were of better quality than orchardgrass silages. When the alfalfa did make poor silage the anaerobic sporeformers occurred later in the fermentation process than in the orchardgrass silages and the numbers obtained were lower.

The increase in numbers of bacteria showed similar trends in all silages. Most of the fresh forages contained relatively few; however, after a few days in the silos the numbers reached several hundred millions per gram of silage, then leveled off and decreased slightly as the silages were held.

The lactic acid bacteria reached about the same maximum in both good and poor quality silages and the few forages that showed high initial numbers of lactic acid bacteria were apparently no better in quality than those with few or none.

The difference in quality of the silages in respect to the occurrence of microorganisms was directly associated with the increase of sporeforming anaerobes. All of the poor quality silages showed substantial increases in these organisms.

As would be expected the good quality silages produced large quantities of lactic acid along with fair amounts of acetic and succinic acids. The poor quality silages, on the other hand, produced butyric acid at the expense of lactic acid. Acetic, propionic and succinic acids occurred along with butyric acid.

The microorganisms isolated from the silages were grouped on the basis of several tests. Representative organisms from the various groups are now being studied in detail.

Table 1

TREATMENT OF SILAGES

Treatment	Orchard Grass			Alfalfa		
	First	Second	Third	First	Second	Third
	Cutting	Cutting	Cutting	Cutting	Cutting	Cutting
1. 20 lbs. sugar/ton of forage, tramped, weighted, sealed immediately.	1 OG 1	2 OG 1	3 OG 1	1 A 1	2 A 1	3 A 1
2. Tramped, weighted, * immediate seal.	1 OG 2	2 OG 2	3 OG 2	1 A 2	2 A 2	3 A 2
3. Tramped, weighted, * sealed after 48 hours.	1 OG 3	2 OG 3	3 OG 3	1 A 3	2 A 3	3 A 3
4. Not tramped, weighted, or sealed.	1 OG 4	2 OG 4	3 OG 4	1 A 4	2 A 4	3 A 4
5. Not tramped, weighted, or sealed, air pumped into bottom of silo for 5-6 hrs.	1 OG 5	2 OG 5	3 OG 5	1 A 5	2 A 5	3 A 5

\* 500 lb. weight.

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Table 2  
CHARACTERISTICS OF THE SILAGES

Good Quality Silages:

- 1) pH values 3.9-4.8.
- 2) Ammoniacal nitrogen values 1.02-2.87%.
- 3) Traces of butyric acid were found in a few of the silages, but most of them contained none.
- 4) Spore counts were erratic; most of the silage contained none.
- 5) Silages showed large amounts of lactic acid 3.03-13.2%.

Poor Quality Silages:

- 1) pH values 5.2-5.7.
- 2) Ammoniacal nitrogen values 3.23-9.82%.
- 3) Spore counts were usually high.
- 4) All silages showed some increase in lactic acid, then a decrease, with a corresponding increase in butyric acid.

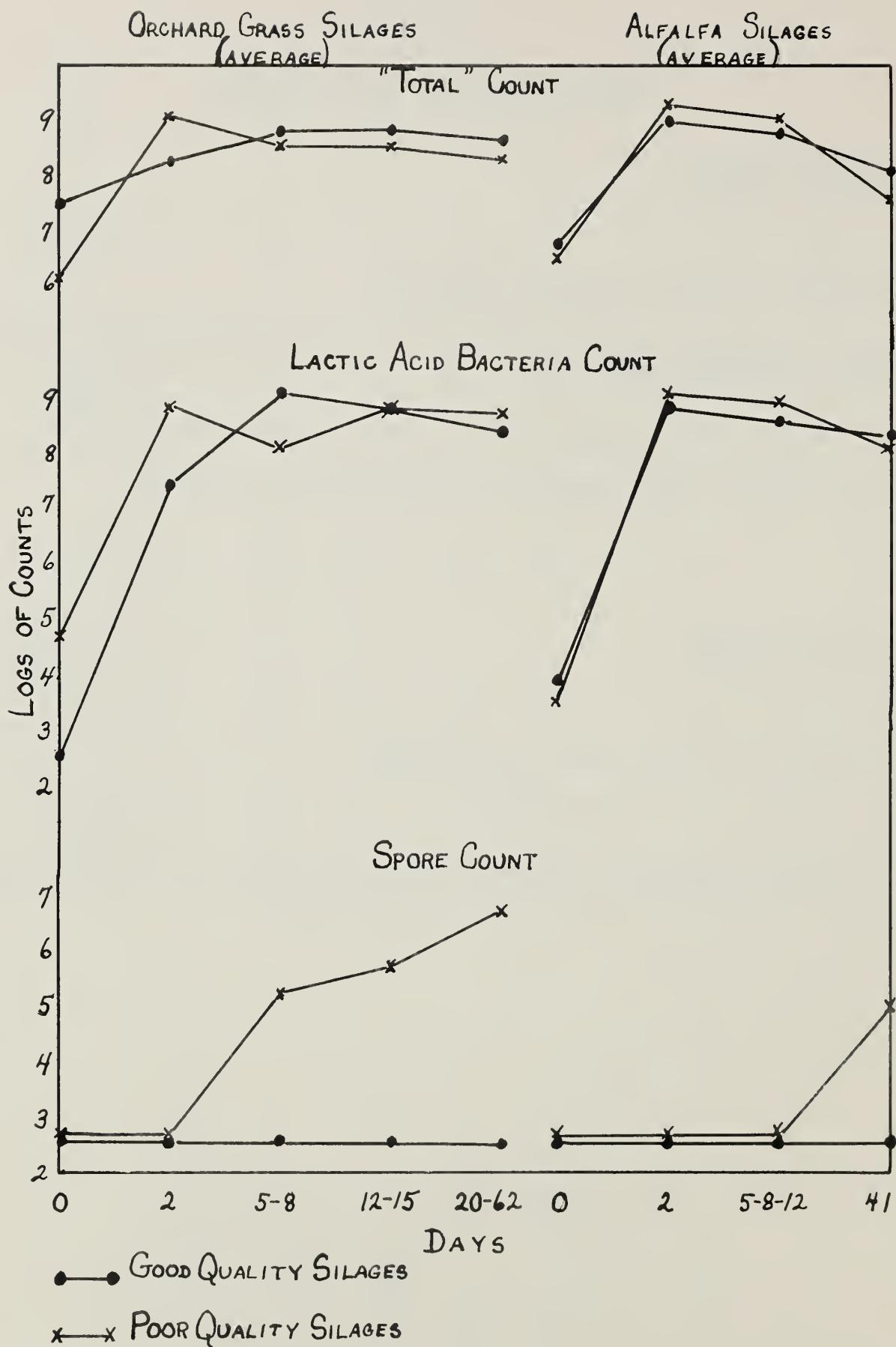


Figure 1. Average numbers of microorganisms in orchardgrass and alfalfa silages.

B BUTYRIC ACID  
P PROPIONIC ACID  
A ACETIC ACID  
F FORMIC ACID  
L LACTIC ACID  
S SUCCINIC ACID

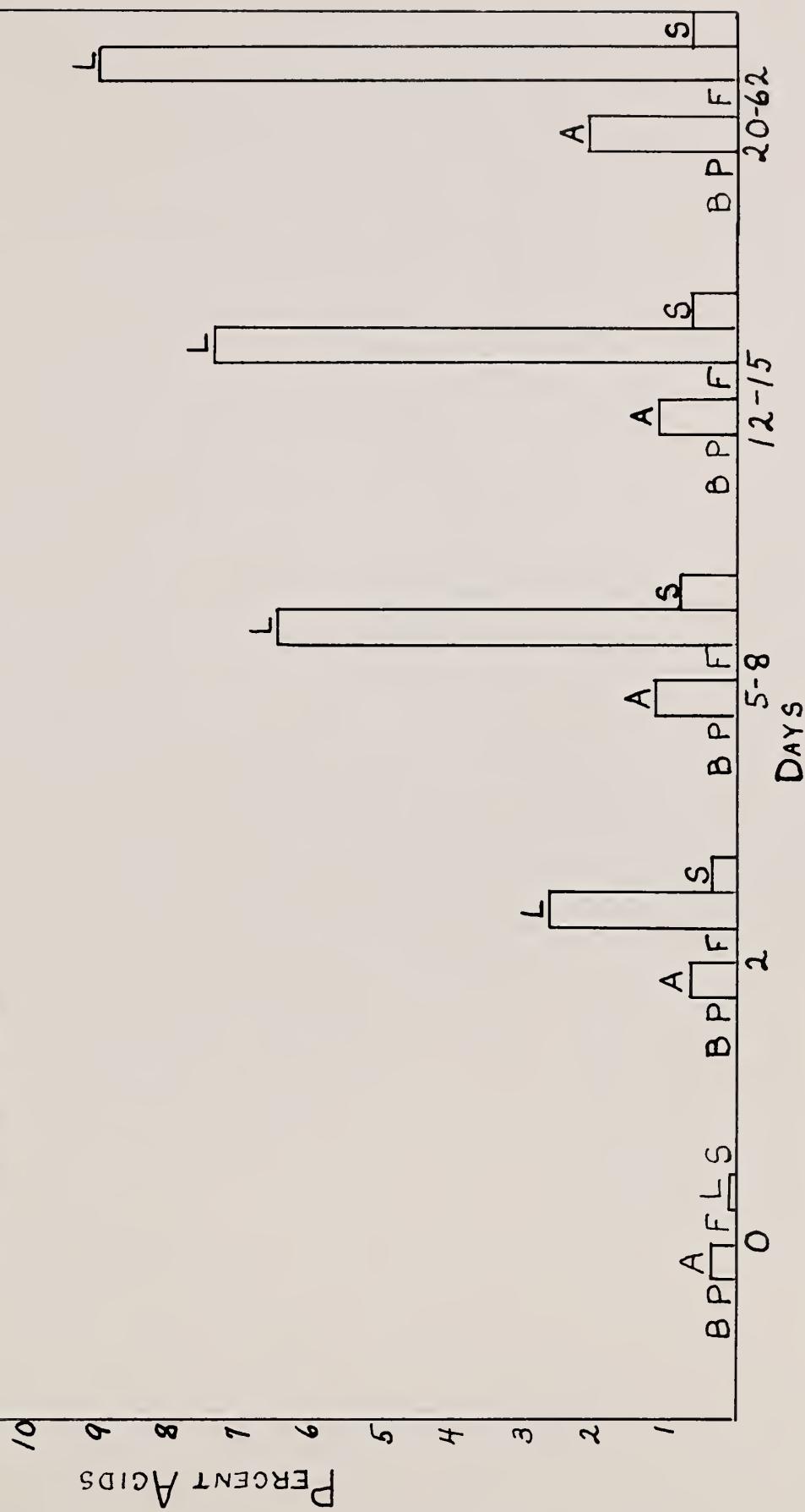


Figure 2. Average amounts of organic acids in good quality silages.

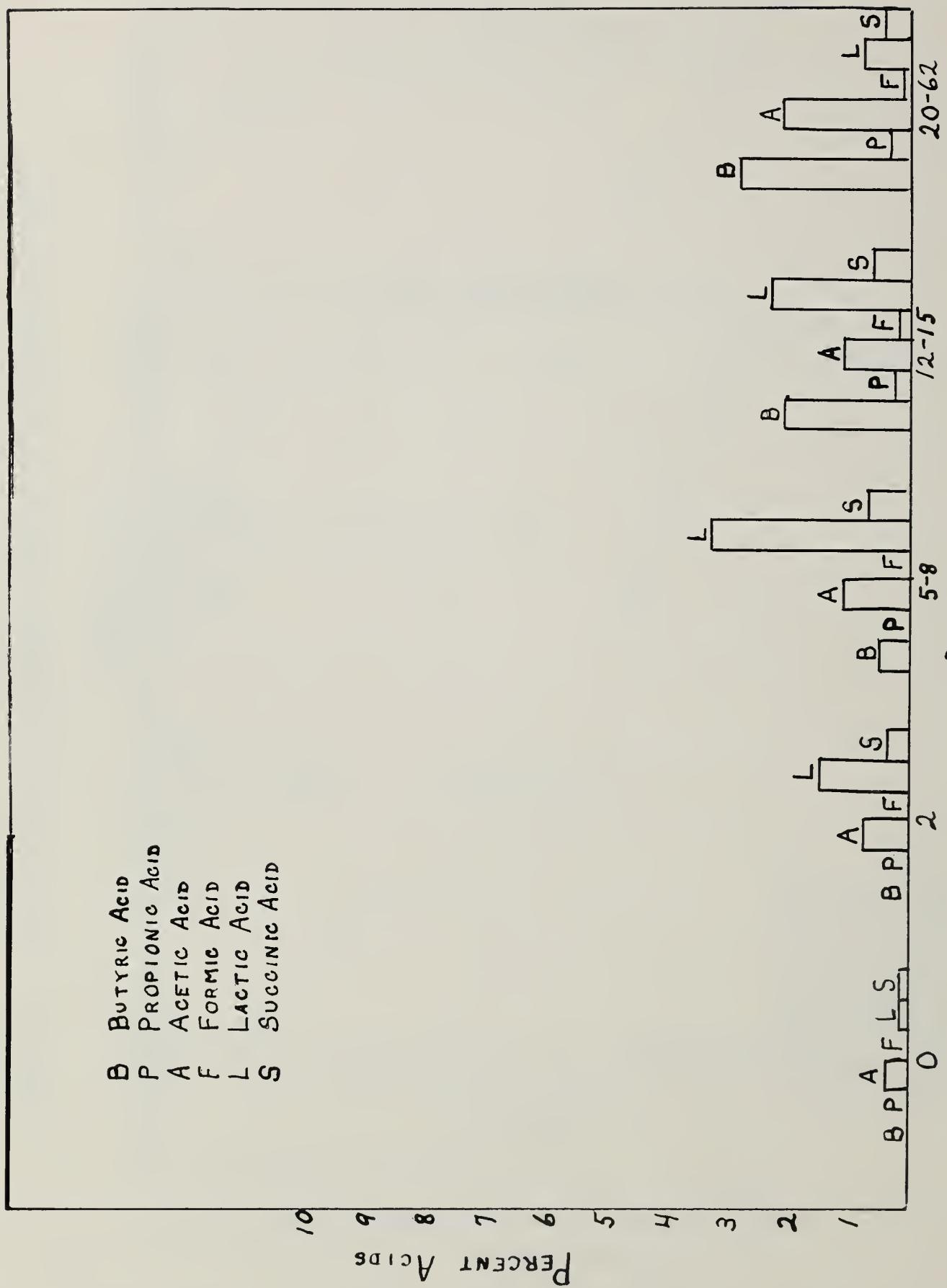


Figure 3. Average amounts of organic acids in poor quality silages.